

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application:

Claims 1-116 and amended claims 1-114 (cancelled).

Listing of Claims

Claim 117 (new): An effective and economical method of processing clinical samples useful for simple, rapid, safe, sensitive diagnosis of bacterial infections such as tuberculosis and other mycobacterial infections caused by mycobacteria including *M. tuberculosis* and other infections caused by Gram-positive organisms like *Staphylococcus sp.* using a composition comprising Solution 1 comprising Universal Sample Processing (USP) solution (composed of Guanidinium Hydrochloride (GuHCl)) of concentration ranging between 3-6 M, Tris-Cl of concentration ranging between 40-60 mM of pH ranging between 7.3-7.7, EDTA of concentration ranging between 20-30 mM, Sarcosyl of concentration ranging between 0.3-0.8%, and beta-mercaptoethanol of concentration ranging between 0.1-0.3 M; Solution 2 selected from the group consisting of a) sodium phosphate of concentration ranging between 65 to 70 mM of pH ranging between 6.7 to 6.8; and b) sterile water; and one or more of Solution 3 comprising Tween 80 of concentration ranging between 0.03 to 0.08%, Solution A comprising Chelex 100 suspension of concentration ranging between 8-12%, Solution B comprising Triton X-100 of concentration ranging between 0.02 to 0.04%, or Solution C comprising Tween 20 of concentration ranging between 0.2-0.4% for isolating DNA, said method comprising steps of:

- (a) obtaining the clinical sample,
- (b) mixing 1.5 to 2 volumes of Solution 1 to the sample,
- (c) homogenizing the mixing while avoiding frothing,
- (d) adding Solution 2 to the homogenate followed by centrifugation to obtain pellet,
- (e) washing the pellet with Solution 1, optionally depending upon the decrease of the pellet size,
- (f) washing the Solution 1-washed pellet with water, and

(g) resuspending the water-washed pellet in one or more of Solution 3, Solution A, Solution B, and/or Solution C to obtain processed sample for diagnosis, the processed sample being used in the form of smear, culture, or Polymerase chain reaction (PCR) starting material, using PCR amplifiable mycobacterial DNA, and RNA.

Claim 118 (new): The method as claimed in claim 117, wherein homogenizing for time duration ranging between 20-120 seconds.

Claim 119 (new): The method as claimed in claim 117, wherein the processing is completed in a total time duration ranging between 1-2 hours.

Claim 120 (new): The method as claimed in claim 117, wherein Guanidinium Hydrochloride is present in a concentration of about 4 M and beta-mercaptoethanol in a concentration of about 0.1 M in the Universal Sample Processing (USP) solution for processing samples for culture and smear, said Guanidinium Hydrochloride being present in a concentration of about 5 M and said beta-mercaptoethanol in a concentration of between 0.1–0.2M for processing of samples for culture, smear and PCR, and said Guanidinium Hydrochloride being present in a concentration of 6 M and said beta-mercaptoethanol present in a concentration of about 0.2 M for samples processed for smear and PCR.

Claim 121 (new): The method as claimed in claim 117 wherein the principal inhibitor removal component in case of specimens containing blood and other inhibitory components is GuHCl.

Claim 122 (new): The method as claimed in claim 117 wherein the principal decontaminating agents are GuHCl and sarcosyl.

Claim 123 (new): The method as claimed in claim 117 wherein the principal mucolytic agent is beta-mercaptoethanol.

Claim 124 (new): The method as claimed in claim 117, wherein PCR-amplifiable mycobacterial DNA and RNA can be obtained through simple lysis by boiling in presence of Solution 3 or by adding 0.01-0.1% Triton X 100 without using Solution A, B and C in case of high bacillary load and/or lesser amount of junk containing samples.

Claim 125 (new): The method as claimed in claim 117, wherein said method in culture runs at a neutral pH.

Claim 126 (new): The method as claimed in claim 117, wherein samples are stored at about -20°C for up to 2 months and can be processed for PCR, smear-microscopy and culture.

Claim 127 (new): The method as claimed in claim 117, wherein said method in PCR uses two sets of primers namely, devRf2 and devRr2, devRf3, and devRr3 of gene *devR* of microbe *Mycobacterium tuberculosis*.

Claim 128 (new): The method as claimed in claim 125, wherein the primers devRf2 and devRr2 amplify a 308 bp fragment of gene *devR* of microbe *Mycobacterium tuberculosis*.

Claim 129 (new): The method as claimed in claim 125, wherein the primers devRf3, and devRr3 amplify a 164 bp fragment of gene *devR* of microbe *Mycobacterium tuberculosis*.

Claim 130 (new): A kit useful in processing clinical samples for simple, rapid, safe, sensitive, and accurate diagnosis of bacterial infections such as tuberculosis and other mycobacterial infections caused by mycobacteria including *M. tuberculosis* and other infections caused by (Gram-positive organisms like *Staphylococcus sp.*,) said kit comprising solution 1 consisting of Universal Sample Processing (USP) solution (composed of Guanidinium Hydrochloride (GuHCl) of concentration ranging between 3-6 M, Tris-Cl of concentration ranging between 40-60 mM of pH ranging between 7.3-7.7, EDTA of

concentration ranging between 20-30 mM, Sarcosyl of concentration ranging between 0.3-0.8%, beta-mercaptoethanol of concentration ranging between 0.1-0.3 M), Solution 2 consisting of Sodium phosphate of concentration ranging between 65 to 70 mM of pH ranging between 6.7 to 6.8 (optionally can be replaced with water), Solution 3 consisting of Tween 80 of concentration ranging between 0.03 to 0.08%, Solution A comprising of Chelex 100 suspension of concentration ranging between 8-12%, Solution B consisting of Triton X-100 of concentration ranging between 0.02-0.04%, and Solution C consisting of Tween 20 of concentration ranging between 0.2 to 0.4, optionally two sets of primers with devRf2 and devRr2 of SEQ ID No. 1 and SEQ ID No. 2 respectively, and primers devRf3, and devRr3 of SEQ ID No. 3, and SEQ ID No. 4 respectively.

Claim 131 (new): The kit as claimed in claim 128, wherein Guanidinium Hydrochloride is present in a concentration of about 4 M and beta-mercaptoethanol in a concentration of about 0.1 M in the Universal Sample Processing (USP) solution for processing samples culture for smear, said Guanidinium Hydrochloride being present in a concentration of about 5 M and said beta-mercaptoethanol in a concentration of between 0.1–0.2 M for processing of samples for culture, smear and PCR, and said Guanidinium Hydrochloride being present in a concentration of 6 M and said beta-mercaptoethanol present in a concentration of about 0.2 M for samples processed for smear and PCR.

Claim 132 (new): The kit as claimed in claim 128, wherein the composition comprises solution 1 consisting of Universal Sample Processing (USP) solution, Solution 2 consisting of Sodium phosphate of concentration ranging between 65 to 70 mM of pH ranging 6.7 to 6.8, Solution 3 consisting Tween 80 of concentration ranging between 0.03 to 0.08%, Solution A comprising Chelex 100 suspension of concentration ranging between 8-12%, and Solution B consisting of Triton X-100 of concentration ranging between 0.02-0.04%, and Solution C consisting of Tween 20 of concentration ranging between 0.2-0.4%.

Claim 133 (new): A set of primers devRf2 and devRr2 of SEQ ID No. 1 and SEQ ID No. 2 respectively.

Claim 134 (new): A set of primers devRf3, and devRr3 of SEQ ID No. 3, and SEQ ID No. 4 respectively.

Claim 135 (new): A method of using primers of SEQ ID No.1, and 2 or SEQ ID No. 3 and 4 of gene *devR* for screening patients of tuberculosis said method comprising steps of conducting Polymerase Chain Reaction (PCR) using DNA or RNA of the processed sample of the subject, identifying the subjects suffering from tuberculosis.